

What is claimed:

1. A packaging cell line for the production of adenovirus, wherein the cell line is capable of producing adenovirus that expresses the A subunit of diphtheria toxin (DT-A) or Pseudomonas Exotoxin A (PEA), and wherein the cell line does not produce replication-competent adenovirus when used in conjunction with non-overlapping E1-deleted adenovirus.
2. The packaging cell line of claim 1, wherein the EF-2 gene in the cell line is mutated.
3. The packaging cell line of any one of claims 1 or 2, wherein the mutated EF-2 gene encodes an EF-2 protein that is mutated at codon 705.
4. The packaging cell line of claim 3, wherein the glycine residue at codon 705 of the EF-2 protein is mutated to arginine.
5. The packaging cell line of any one of claims 1-4, wherein the cells are resistant to about 10^{-9} M diphtheria toxin.
6. The packaging cell line of any one of claims 1-5, wherein the cells contain the adenovirus E1 region.
7. The packaging cell line of any one of claims 1-6, wherein the cells contain the adenovirus serotype 5 (Ad5) E1-A and E1-B encoding sequences.
8. The packaging cell line of any one of claims 1-7, wherein the cells are derived from PER.C6 cells.
9. A method of producing adenovirus which expresses the A subunit of diphtheria toxin (DT-A) or Pseudomonas Exdotoxin A (PEA), wherein the method does not produce replication-competent adenovirus, comprising:
 - a) infecting the packaging cell line of any one of claims 1-8 with non-overlapping E1-deleted adenovirus which expresses DT-A or PEA; and
 - b) culturing the cells for an amount of time sufficient to produce adenovirus.

10. The method of claim 9, further comprising isolating adenovirus from the cells.

11. The method of any of claims 9 or 10, further comprising isolating adenovirus
5 from the culture medium.

12. The method of any of claims 9-11, wherein expression of the DT-A or PEA is under the control of a tissue-specific promoter or enhancer.

10 13. The method of claim 12, wherein the tissue-specific promoter or enhancer is a prostate-specific promoter or enhancer.

14. The method of claim 13, wherein the prostate-specific promoter or enhancer comprises the first five kilobases upstream of the transcription start site of the prostate-specific antigen (PSA) gene.
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15. Adenovirus produced by the method of any one of claims 9-14.

16. A method of killing a cell that is sensitive to DT-A or PEA, comprising
20 infecting the cell with the adenovirus of claim 15.

17. The method of claim 16, wherein the cell is a cancer cell.

18. The method of any of claims 16 or 17, wherein the cell is a prostate cancer
25 cell.

19. A method of selectively killing a cell in a subject, comprising administering a therapeutically effective amount of a the adenovirus of claim 16 to the subject, wherein the tissue-specific promoter or enhancer that controls the expression of the DT-A or PEA is
30 active only in the cell and not in other cells, thereby killing the cell but not other cells.

20. The method of claim 19, wherein the cell is a cancer cell.

21. The method of claim 20, wherein the cell is a prostate cancer cell.
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22. A method of treating a subject suffering from cancer comprising administering a therapeutically effective amount of the adenovirus of claim 15 to the subject, thereby treating said cancer.

23. The method of claim 22, wherein the cancer is prostate cancer.

24. A method of producing an immunotoxin comprising diphtheria toxin A or
5 Pseudomonas exotoxin A, comprising:

- a) contacting the packaging cell line of any one of claims 1-8 with a nucleic acid molecule which encodes the immunotoxin; and
- b) culturing the cells for an amount of time sufficient to produce the immunotoxin.

10 25. The method of claim 24, further comprising isolating the immunotoxin from the cells.

15 26. The method of any one of claims 24 or 25, further comprising isolating the immunotoxin from the culture medium.

27. An immunotoxin produced by the method of any one of claims 25-26.

20 28. A method of making a cell resistant to diphtheria toxin or Pseudomonas Exotoxin A comprising:

a) contacting a cell which is sensitive to diphtheria toxin or Pseudomonas Exotoxin A with a nucleic acid molecule encoding a fragment of the EF-2 protein, wherein the fragment comprises a mutation at codon 705;

25 b) culturing the cell for a period of time sufficient to allow homologous recombination to occur between the nucleic acid molecule and the endogenous EF-2 gene; and

c) contacting the cell with an amount of diphtheria toxin or Pseudomonas Exotoxin A sufficient to kill a cell which is not resistant to diphtheria toxin or Pseudomonas Exotoxin A,

30 wherein growth or division of the cell in the presence of diphtheria toxin or Pseudomonas Exotoxin A indicates that the cell has been made resistant to diphtheria toxin or Pseudomonas Exotoxin A.

35 29. The method of claim 28, wherein the mutation at codon 705 of the EF-2 protein comprises a mutation from glycine to arginine.

30. The method of any of claims 28 or 29, wherein the nucleic acid molecule is less than about 500 nucleotides in length.

31. The method of any of claims 28 or 29, wherein the nucleic acid is about 500 nucleotides in length.

5 32. The method of any of claims 28 or 29, wherein the nucleic acid molecule is less than about 500 nucleotides.